Synergistic Effects of Biofertilizers and Antioxidants on Growth and Nutrients Content of Corn under Salinity and Water-Deficit Stresses

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ABSTRACT

A greenhouse pot experiment was conducted to explore ways to alleviate the deleterious effects of saline-irrigation water on the growth and the yield of corn, Water with three salinity levels was applied to soil, tap water (0.47), 2.50, and 3.90 dSm^{-1}. As a biofertilizer, "Halix" containing *Azotobacter, Azospirillum, and Klebsiella* species was applied as an inoculum to corn seeds before cultivation. Ascorbic acid and mannitol as antioxidants (100 mg l^{-1}) were added as a foliar spray. Data showed that all growth parameters such as, contents of total Chlorophyll, ascorbic acid, micro- and macro-nutrients, and corn dry weights responded negatively as the salinity level increased. Results showed that the combined treatments of biofertilizer and ascorbic acid have significantly alleviated the adverse effects of salinity on corn growth performance. The combined treatments of biofertilizer and ascorbic acid significantly increased macro and micro-nutrients concentration, total chlorophyll, and ascorbic acid contents in corn plants compared to the untreated and salinity affected plants. In general, the increase of salinity levels significantly increased proline content in corn plants. Among all treatments, it can be concluded that the combination of mycorrhiza and plant growth promoting rhizobacteria (PGPR) as biofertilizers and ascorbic acid as an antioxidant treatments has a promising effect for alleviation of adverse effects of salinity on growth performance of corn plant.

Key Words: **Antioxidant, biofertilizer, drought, salinity, mycorrhiza, PGPR, corn**

INTRODUCTION

It is well established that drought and salt affected soils have adverse effects on most field crops,therefore, plants grown under salt or water-deficit stresses use a variety of strategies to counteract and relief these oxidative stress damages. Among these strategies, ion homeostasis, enhanced antioxidant activity, and photosynthetic capacity. Many investigators showed that salt tolerance in most crops is associated with a more efficient antioxidant system (Gossett et al., 1994, 1996; Noctor and Foyer, 1998; Mittova et al., 2002; Bor et al., 2003). An efficient antioxidant system includes enzymatic (superoxide dismutase, SOD; catalase, CAT; ascorbate peroxidase, APX; glutathione reductase, GR; dehydroascorbate reductase, DHAR; glutathione peroxidase, GPX; and glutathione-S- transferase, GST), and non-enzymatic antioxidants (ascorbic acid, AsA; glutathione, GSH; phenolic compounds, alkaloids, tocopherols, salicylic acid, and carotenoids) (. Ascorbic acid (AsA) is one of the most important antioxidants protecting plants from oxidative stress (Smirnoff, 2005). Ascorbic acid and mannitol were shown also to be involved in regulating photosynthetic capacity, flowering and senescence (Davey et al., 2000), and in counteracting adverse effects of salt and water-deficit stresses in tomato (Shalata and Neumann, 2001) and in wheat (Al-Hakimi and Hamada, 2001).

Biofertilizers are products containing living cells of different types of microorganisms, which have an ability to convert nutritionally important elements from unavailable to available form through biological processes (Hegde et al., 1999; Vessey, 2003). In recent years, biofertilizers have emerged as an important component of the integrated nutrient supply system and hold a great promise to improve crop yields through environmentally better nutrient supplies than conventionally common inorganic fertilizers. Some of these soil microorganisms are effective on plant growth and are called plant growth promoting rhizobacteria (PGPR). Halix is one of the biofertilizers used in Egypt containing three species of free nitrogen fixers; *Azospirilla, Azotobacter, and Klebsiella*. These three species are also among plant growth promoting rhizobacteria (PGPR) which provide other functions beside their main recognized influence as free nitrogen fixers (Gray and Smith 2005). PGPR that can exert a positive plant growth by direct mechanisms such as solubilization of nutrients, nitrogen fixation, production of growth regulators, etc., or indirectly such as stimulation of mycorrhiza development, competitive exclusion of pathogens or removal of phytotoxic substances (Bashan and de-Bashan 2010). Bacilio et al. (2004) found that, under high NaCl concentration, inoculation with modified *A. lipoferum* reduced the deleterious effects of NaCl.

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Mycorrhiza is another example of biofertilizers,hence, it showed a positive effect of protecting plants from salt and drought damages. Inoculation with Halix and Mycorrhiza under saline and water-deficit stresses conditions is useful as it can accumulate compatible solutes, such as glycine, betaine, glutamate, and proline, causing adaptation to fluctuations in soil salinity. Asghari (2008) found that the enhancement of clover dry weight resulting from mycorrhiza inoculation was greater under high salinity levels. He also reported that the detrimental effects of salinity stress on plant growth were appeared immediately after application of low salinity stress to nonmycorrhizal plants (3.5 dS/m), but it was only observed in mycorrhiza inoculated plants at 7.5 dS/m and above. The objective of the present work was to explore the effects of saline and drought stresses on yield of corn plants, as well as to mitigate the physiological and biochemical impacts induced by salinity and water-deficit stresses when bio-fertilization and antioxidant were applied.

MATERIALS AND METHODS

Soil characteristics:

A surface soil layer (0-20 cm) was collected from Almowazafeen Village at Abis, Alexandria, Egypt. The soil was air-dried and sieved through a 2-mm sieve. Soil pH and electrical conductivity (EC) were determined in soil-paste extract (Richards, 1954), organic matter content was determined by dichromate oxidation method (Nelson and Sommers, 1982), cation exchange capacity (CEC) was determined by 1M NaOAC method (Rhoades, 1982), particle size distribution was determined by the hydrometer method (Day, 1965), total calcium carbonate content was determined using calcimeter (Nelson, 1982), available phosphorus was determined by 0.5 M $NaHCO_3$ test (Olsen and Sommers, 1982), available nitrogen was determined by 2M KCl extraction method (Bremner and Mulvaney, 1982), and available potassium was determined by ammonium acetate method (Knudsen and Peterson,1982). The main chemical and physical properties of soil are shown in Table (1).

Corn seeds:

Seeds of corn (*Zea mays)* were a white pioneer single hybride (30K8). It is collected from Ministry of Agriculture and Land Reclamation (MALR), Egypt.

Experimental procedure:

Seven kilograms of sandy clay loam soil were weighed in polyethylene pots (diameter 25 cm), then irrigated with water of different salinity. The EC of each saline water was obtained by blending tap water with sea water thoroughly and then measuring EC. The three salinity levels (S1, S2, and S3) were: 0.47 (tap water), 2.50, and 3.90 dSm^{-1} representing 100, 75, and 50 % of corn yield, respectively (FAO, 1976).

The water-deficit treatments were 100, 75, and 50% from water holding capacity (WHC) of soil.

The Biofertilizer" Halix" was applied as an inoculum to corn seeds before cultivation. Mycorrhiza inoculum was prepared by soil enrichment technique using corn as a host for vesicular arbuscular mycorrhiza (VAM). The inoculum was analyzed for its content of mycorrhiza spores. This inoculum was found to contain an average 1.5×10^4 spores/gm. 15 gm of this inoculum was homogenously mixed with the upper 10 cm layer of pot after seed germination Philips and Hayman (1970). Pots were placed in greenhouse, receiving only natural light, for 16 weeks, and irrigated weekly with tap water (EC 0.47 dS m^{-1}) and/or saline water levels. Fertilization with the recommended rates of N, P, and K was accomplished according to MALR. The maximum and minimum temperatures in the greenhouse were 35 and 25 ^{0}C, respectively. Ascorbic acid and Mannitol (100 mg l^{-1}) were added as a foliar spray twice after two and four weeks of sowing date. The experimental design was a split plot design with four replicates of each treatment. Plant root and shoot samples were taken after 60 days from planting and dried at 70 °C in oven for 48 hrs and weighed.

Plant sampling and analysis:

Samples of plants were collected after seven weeks from sowing for chemical analysis of leaves except for total chlorophyll which was analysed in the upper third leaf of corn plants of all treatments. These plant samples were immediately rinsed three times in distilled water to remove the adhering soil and dust particles. Oven-dried plant materials were grounded in a stainless steel mill and subsamples were dry-ashed in a muffle furnace at 450 °C for 6 h. Ash was dissolved in 5 ml of HNO_3 (1: 1), diluted to a constant volume with distilled water and analysed for K, P, Fe, Mn, Zn, Cu, Na, and Cl (Jones, 2001). Another sub-sample was ashed and dissolved in hydrochloric acid solution (1:1, v/v), diluted to a certain volume with double-distilled water, and analysed for N (Jones, 2001) by Kjeldahl method (Bremner and Mulvaney, 1982). The dry matter production and plant nutrients concentrations are expressed on oven-dry weight basis. Mycorrhizal colonization was observed by taking random root samples from every treatment and subjected to clearing and staining method with Trypan Blue stain (Philips and Hayman, 1970). Not less than 30 root pieces was observed under the microscope for infection percentage determination (Giovannetti and Mosse 1980).

Table 1. Physio-chemical characteristics of used soil

Soil characteristic	Unit	Value[¶]
pH (H_2O)		8.08±0.09
EC	dSm^{-1}	2.11±0.34
CEC	$Cmol(+).kg^{-1}$	27.26±1.45
$CaCO_3$	%	5.20±0.33
OC	%	1.19±0.09
OM	%	2.05±0.08
Available-N	mg. kg^{-1}	14.24±0.89
Olsen-P	mg. kg^{-1}	15.55±0.43
Available-K	mg. kg^{-1}	132.85± 6.43
Clay	%	34.30± 0.92
Silt	%	10.40±0.42
Sand	%	55.30± 5.87
Texture		S.C.L[¶¶]
W.H.C[††]	%	30

[¶] Means of three samples ± SD.

[†] O.M: organic matter; [¶¶] S.C.L: sandy clay loam,

[††] W.H.C.: Water holding capacity,

Chemical constituents:

Seven weeks after planting, the third leaf of plant was collected from each pot for chemical analyses. Chlorophyll was extracted from leaves by 80 % acetone, and then determined (mg g^{-1} FW) using a colorimetric method (Arnon 1949). Free proline was extracted by sulfosalicylic acid (3%) then determined colorimetrically (mg g^{-1} DW) using acid ninhydrin reagent as outlined by Bates et al., (1973). Ascorbic acid was determined (mg g^{-1} FW) according to AOAC (1995).

Statistical analysis:

The treatment effects on growth and yield parameters, chemical constituents, and nutrients concentrations were evaluated by analysis of variance (ANOVA) and also by the least significant difference (LSD) mean separation procedure at 0.05 level of significance (SAS Institute, 1994).

RESULTS AND DISCUSSION

Dry matter yield of corn plants affected by water and salinity stresses:

The effects of salinity and water stresses levels on dry matter yields of corn plants are presented in Table (2). In general, the addition of biofertilizer and antioxidants significantly mitigated the negative effects of salinity and water-deficit treatments. It was found that application of biofertilizers (H+MR) significantly increased the yield of corn above that of the untreated plants. Also, these data showed that ascorbic acid application (AsA) significantly increased dry matter yield of corn plants in comparison to control. Moreover, the combined treatments of biofertilizer and antioxidants levels (H+MR+AsA+) and (H+MR+M) significantly ameliorate the negative effects of salinity levels on corn dry matter yields. among all, treatments of ascorbic acid combined with the two biofertilizers (H+MR+AsA) have shown a synergistic effect protecting corn plants from the adverse effect of salinity through a significant increase of its yield followed by mannitol and both biofertilizers (H+MR+M) (Table 2).

Similarly, Table (2) showed that the addition of both biofertilizers (H+MR) and ascorbic acid rates (AsA) significantly increased dry weight of corn plants. Moreover, the combined treatments of both biofertilizers, antioxidant levels (H+MR+AsA) and (H+MR+M) significantly ameliorated the negative effects of water-deficit levels on corn dry matter yields. Although both antioxidants improved the yield of corn,however ascorbic acid treatment (H+MR+AsA) showed a better effect than mannitol (H+MR+M) in protecting corn plants against the adverse effects of salinity and drought (Table 2). Also, adding of 75% of water holding capacity gave the highest yield of corn.

This decrease in corn yield could be explained in the basis that salinity has both osmotic and specific ion effects on plant growth (Dionisio-Sese and Tobita 2000). However, many workers showed that the deleterious effect of salinity or water-deficit was attributed to salt induced stress, ion toxicity, ion imbalance or acombination of all of these factors (Kurt et al., 1986). Salinity and water-deficit stresses reduced both of shoot and root dry weights of plants (Hamdy et al. 1993; Essa 2002; Li et al.2006; Sharifi et al., 2007; Mahdy and Fathi, 2012).

Table 2. Effects of salinity and biofertilizer treatments on dry matter yield of corn plants

Salinity and water stresses treatments	Amendments	Dry matter yield, g	
		Shoot	Root
S1	Control	65.00±4.52	13.22±1.12
	H+MR	5.33±77.54	2.55±18.12
	AsA	4.98±72.48	3.24±15.87
	M	6.12±70.22	2.66±14.11
	H+MR +AsA	10.58±121.12	3.33±20.36
	H+MR +M	9.65±115.24	2.45±18.87
LSD $_{0.05}$		**3.45**	**1.45**
S2	Control	5.24±54.74	1.11±8.98
	H+MR	5.11±61.33	1.02±11.23
	AsA	4.25±59.14	1.24±10.22
	M	3.87±59.00	0.88±9.87
	H+MR +AsA	5.12±66.05	1.22±14.00
	H+MR +M	6.35±64.11	1.23±12.54
LSD $_{0.05}$		**1.88**	**1.07**
S3	Control	4.88±45.87	0.78±6.21
	H+MR	3.44±53.78	1.57±8.65
	AsA	4.22±51.12	0.88±7.51
	M	5.32±50.24	0.57±6.87
	H+MR +AsA	4.67±60.88	2.54±11.38
	H+MR +M	5.32±58.91	1.54±10.68
LSD $_{0.05}$		**1.22**	**0.88**
100% W.H.C.	Control	3.87±60.22	2.00±11.35
	H+MR	68.23±5.12	15.32±1.18
	AsA	6.21±65.89	1.88±12.33
	M	3.22±63.25	0.99±10.87
	H+MR +AsA	10.21±110	2.11±17.44
	H+MR +M	9.88±105.32	1.79±16.22
LSD $_{0.05}$		**1.35**	**1.01**
75% W.H.C.	Control	65.00±4.52	13.22±1.12
	H+MR	5.33±77.54	2.55±18.12
	AsA	4.98±72.48	3.24±15.87
	M	6.12±70.22	2.66±14.11
	H+MR +AsA	10.58±121.12	3.33±20.36
	H+MR +M	9.65±115.24	2.45±18.87
LSD $_{0.05}$		**3.45**	**1.45**
50% W.H.C.	Control	3.88±50.87	1.09±8.11
	H+MR	3.65±59.66	1.33±11.57
	AsA	4.02±56.99	1.55±9.88
	M	5.55±54.32	0.98±8.11
	H+MR +AsA	9.54±85.87	1.85±14.57
	H+MR +M	8.88±81.98	2.12±12.35
LSD $_{0.05}$		**1.74**	**0.93**

H+MR: Halix + Mycorrhiza AsA: Ascorbic acid M: Mannitol

¶ Means of four samples ± SD

Rengasamy (2002) showed that the reduction in plant growth under saline condition may be due to low osmotic potentials resulting from salinity that can restrain water uptake by plants, which reduces their ability to survive and produce. Dry matter yield reduction due to salinity was explained by various workers (Greenway and Munns 1980; Schwarz and Gale 1981; Walker et al., 1981). They stated that salinity causes physiological and biochemical disorders in plants. Also, it is reported that biofertilizers similar to "Halix and Mycorrhiza" are capable of positively affecting the growth and yield of numerous plant species, as it is capable of producing various phytohormones that improve plant growth (Bashan et al., 2004).

Total chlorophyll and chemical constituents Contents:

The data presented in Table (3) showed that salinity and/or water-deficit treatments significantly decreased total chlorophyll contents in leaves of corn plants compared to that of the control. It is very obvious that both biofertilizer treatments (H+MR) significantly protected corn plants against the reduction in total chlorophyll content occurred at high salinity levels (Table 3). Hence, application of (H+MR) stimulated chlorophyll synthesis under salt stress. Data also showed that ascorbic acid or mannitol application (100 mg l^{-1}) mitigated the negative impact of low salinity level, 2.50 dSm^{-1} (S2), on total chlorophyll content, while slightly reduced the negative effect at a higher salinity level, 3.90 dSm^{-1}(S3). On the other hand, the combined treatments of biofertilizer and ascorbic acid (H+MR+AsA) or mannitol (H+MR+M)) significantly increased total chlorophyll contents in the leaves of corn plants (Table 3).

This could be due to that ascorbic acid and mannitol were used as antioxidants in association with other components of the system. Also, it is stated that ascorbic acid protects plants against oxidative damage resulting from aerobic metabolism, photosynthesis and a range of pollutants and salt and drought stresses(Bashan et al., 2004). While ascorbic acid (vitamin C) is a familiar molecule because of its dietary significance, most aspects of its metabolism and some aspects of its function in plants are very poorly understood(Bashan et al., 2004).

The results in Table (3) showed that ascorbic acid concentration in corn tissues of different treatments, ranged from 0.11 to 0.49 $mg.g^{-1}$and increased with the increase of soil salinity level. Also, data showed that treating soil with either ascorbic acid or mannitol significantly changed the level of ascorbic acid relative to the untreated plants at the three tested salinity levels; however addition of both biofertilizers with either antioxidant has increased AsA significantly in corn plants at both S2 and S3 salinity treatments over single treatments but not the control plants.

Table (3) showed that increasing of salinity significantly increased proline content in corn plants. Biofertilizer treatments (H+MR), (H+MR+AsA), and (H+MR+M) significantly decreased proline content in corn leaves compared with that of the control. Otherwise, there was a significant impact on proline content in corn plants with ascorbic acid application (100 mg l^{-1}) at salinity, 2.50 and 3.90 dSm^{-1} treatments (Table 3). These results coincided with the results of other researchers (Hamdy et al.1993; Essa 2002; Li et al. 2006; Sharifi et al. 2007; Mahdy and Fathi, 2012). Proline has also a dual role in improving salt stress tolerance as it is able to act in a similar way to the peroxidase enzymes and scavenge reactive oxygen species (Zhu, 2001). High levels of free proline evident in corn may have had a protective effect on cells at a higher concentration of salts. Proline is a key osmolyte that contributes to osmotic adjustment and improves stress tolerance by protecting and stabilizing membranes and enzymes during stress conditions.

These data also indicated that the addition of ascorbic acid or mannitol improved the dry weight of corn plants. Consistent findings reported the beneficial effects of exogenous applications of ascorbic acid in partially mitigating the adverse effects of salt stress on plants growth (Mozafar and Dertli 1992). The obtained results revealed a significant interaction effects between NaCl stress and exogenous ascorbic acid on the chlorophyll content, since salt and water-deficit stresses can lead to oxidative stress through the increase in reactive oxygen species (ROS) which are highly reactive and may cause cellular damage. One of the proposed biochemical modes of ascorbate is to act as an antioxidant by scavenging hydrogen peroxide (chloroplasts lack catalase) as it forms (Miyake and Asda 1992).

Nutrients content in corn:

Figures 1, 2, 3 and 4 show macro- and micro-nutrients in corn as affected by all treatments. Figure (1) demonstrates the effects of the tested factors on the elements contents of corn plants. Salinity levels (S2 and S3), showed that N, P and K have significantly decreased with salinity increase, while Na and Cl concentrations were significantly increased. However, S3 treatment is slightly affected.

Table 3. Effects of salinity levels and biofertilizer treatments on total chlorophyll, ascorbic acid, and proline in corn plants

Salinity and water stresses treatments	Amendments	Total chlorophyll, mg g^{-1} FW	Ascorbic acid, mg g^{-1}	Proline, mg g^{-1}
S1	Control	0.18±0.84	0.06±0.24	0.05±0.17
	H+MR	0.21±0.88	0.05±0.20	0.02±0.14
	AsA	0.23±0.95	0.05±0.23	0.03±0.16
	M	0.18±0.93	0.03±0.18	0.05±0.13
	H+MR +AsA	0.17±1.16	0.08±0.27	0.04±0.12
	H+MR +M	0.33±1.05	0.04±0.28	0.02±0.10
LSD 0.05		**0.05**	**0.04**	**0.03**
S2	Control	0.11±0.68	0.08±0.41	0.13±0.45
	H+MR	0.14±0.79	0.07±0.28	0.05±0.38
	AsA	0.17±0.92	0.09±0.32	0.11±0.37
	M	0.84±0.22	0.29±0.08	0.35±0.08
	H+MR +AsA	0.33±1.12	0.11±0.45	0.08±0.31
	H+MR +M	0.96±0.28	0.47±0.08	0.28±0.07
LSD 0.05		**0.08**	**0.09**	**0.04**
S3	Control	0.15±0.48	0.09±0.47	0.10±0.55
	H+MR	0.11±0.58	0.08±0.37	0.08±0.43
	AsA	0.08±0.71	0.09±0.33	0.08±0.42
	M	0.68±0.13	0.27±0.08	0.06±0.39
	H+MR +AsA	0.12±0.86	0.07±0.47	0.09±0.34
	H+MR +M	0.81±0.15	0.11±0.49	0.05±0.32
LSD 0.05		**0.08**	**0.09**	**0.04**
100% W.H.C.	Control	0.17±0.80	0.04±0.20	0.04±0.15
	H+MR	0.24±0.82	0.02±0.16	0.03±0.12
	AsA	0.20±0.88	0.04±0.18	0.03±0.13
	M	0.16±0.83	0.03±0.14	0.06±0.11
	H+MR +AsA	0.11±0.98	0.07±0.23	0.03±0.10
	H+MR +M	0.22±0.96	0.05±0.25	0.02±0.08
LSD 0.05		**0.07**	**0.06**	**0.02**
75% W.H.C.	Control	0.18±0.84	0.06±0.24	0.05±0.17
	H+MR	0.21±0.88	0.05±0.20	0.02±0.14
	AsA	0.23±0.95	0.05±0.23	0.03±0.16
	M	0.18±0.93	0.03±0.18	0.05±0.13
	H+MR +AsA	0.17±1.16	0.08±0.27	0.04±0.12
	H+MR +M	0.33±1.05	0.04±0.28	0.02±0.10
LSD 0.05		**0.05**	**0.04**	**0.03**
50% W.H.C.	Control	0.15±0.50	0.06±0.11	0.09±0.25
	H+MR	0.14±0.63	0.05±0.14	0.09±0.22
	AsA	0.10±0.58	0.03±0.16	0.08±0.23
	M	0.11±0.53	0.03±0.11	0.06±0.21
	H+MR +AsA	0.13±0.78	0.06±0.17	0.02±0.13
	H+MR +M	0.12±0.76	0.08±0.15	0.02±0.10
LSD 0.05		**0.11**	**0.05**	**0.04**

H+MR: Halix + Mycorrhiza AsA: Ascorbic acid M: Mannitol

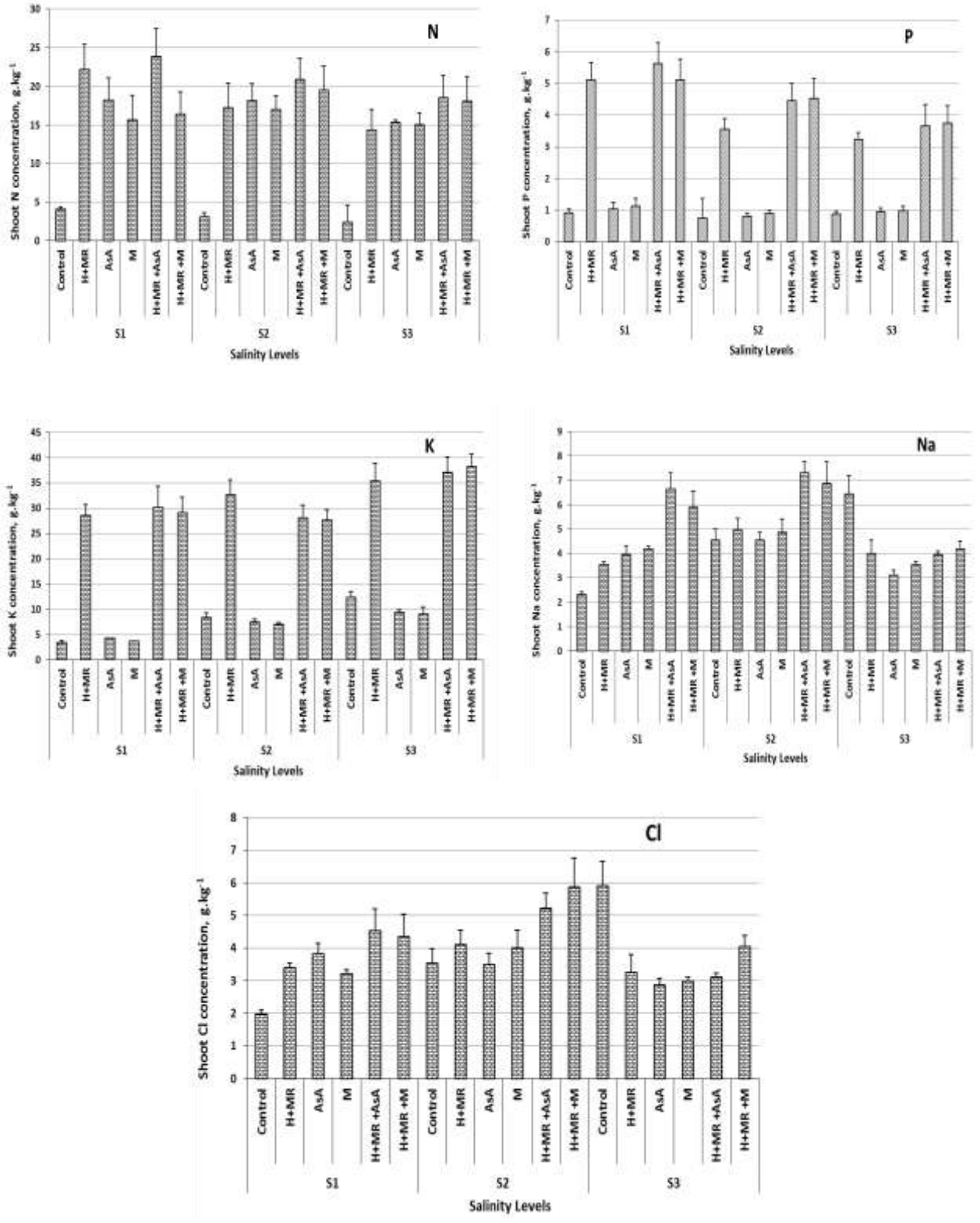

Figure 1. Effects of soil salinity, biofertilization, and antioxidant treatments on N, P, K, Na, and Cl Concentrations in corn. Error bars on all figures represent the standard error of the mean.

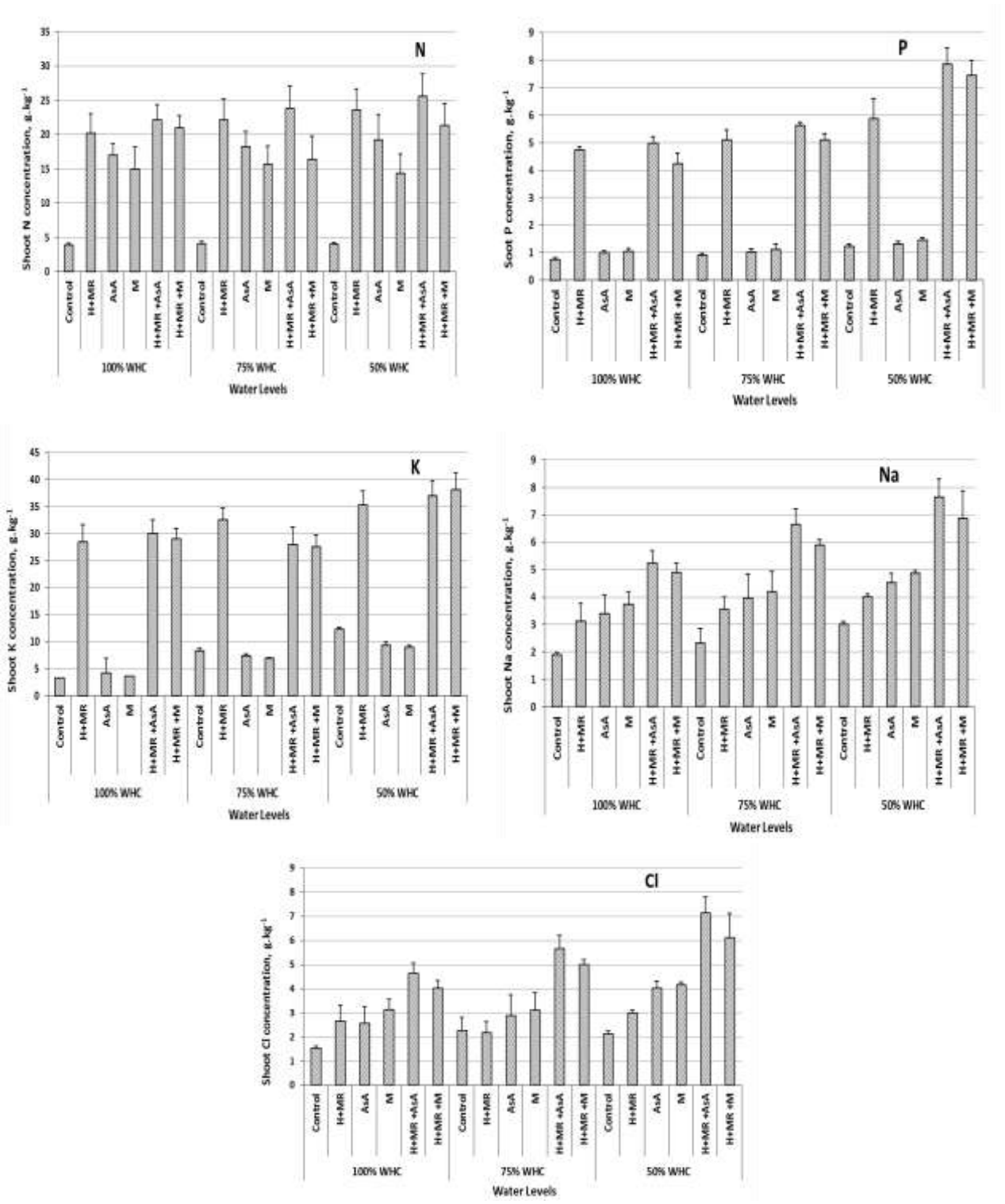

Figure 2. Effects of water stress levels, biofertilization, and antioxidant treatments on N, P, K, Na, and Cl concentrations in corn. Error bars on all figures represent the standard error of the mean.

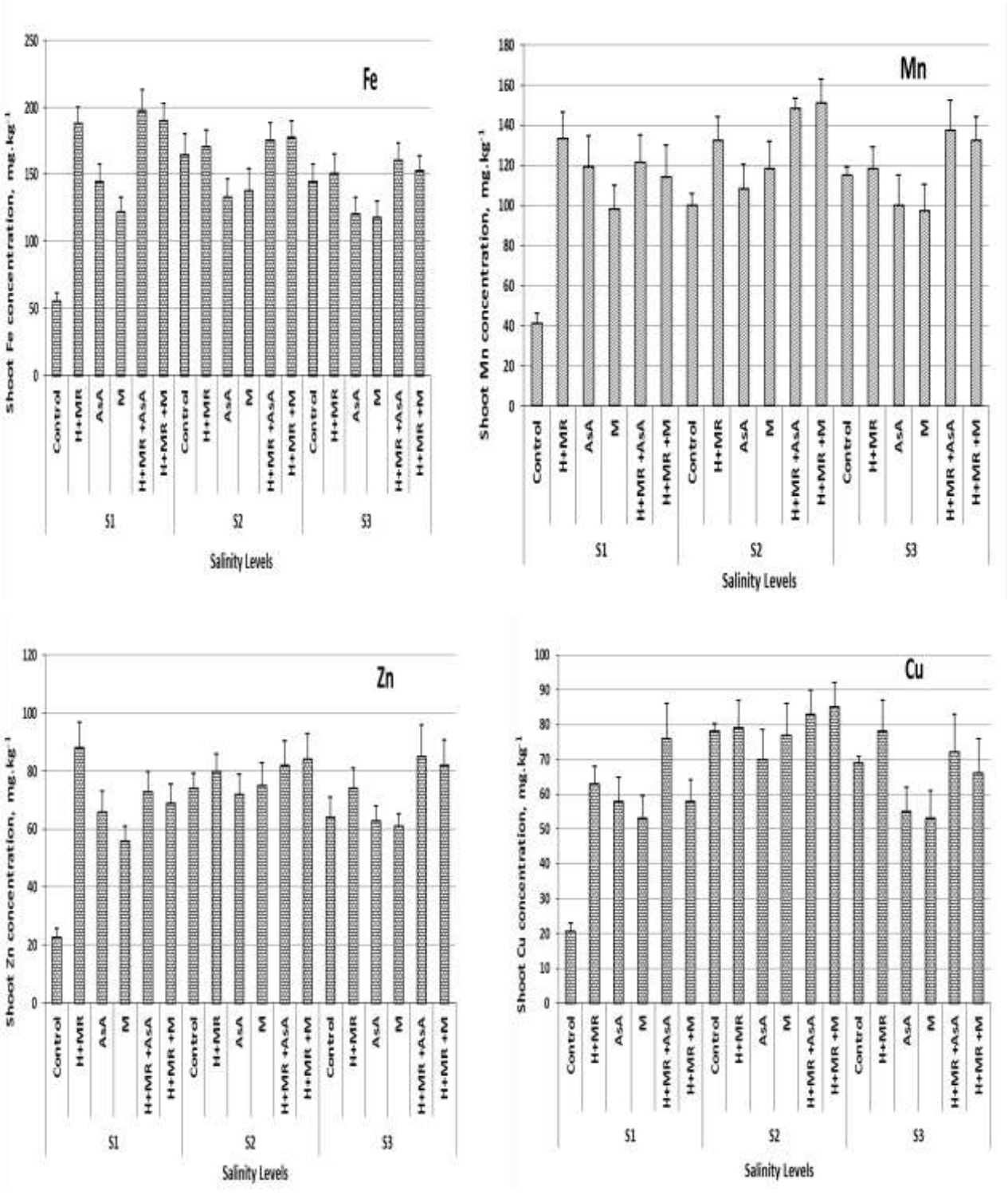

Figure 3. Effects of soil salinity, biofertilization, and antioxidant treatments on some micronutrients concentrations in corn. Error bars on all figures represent the standard error of the mean.

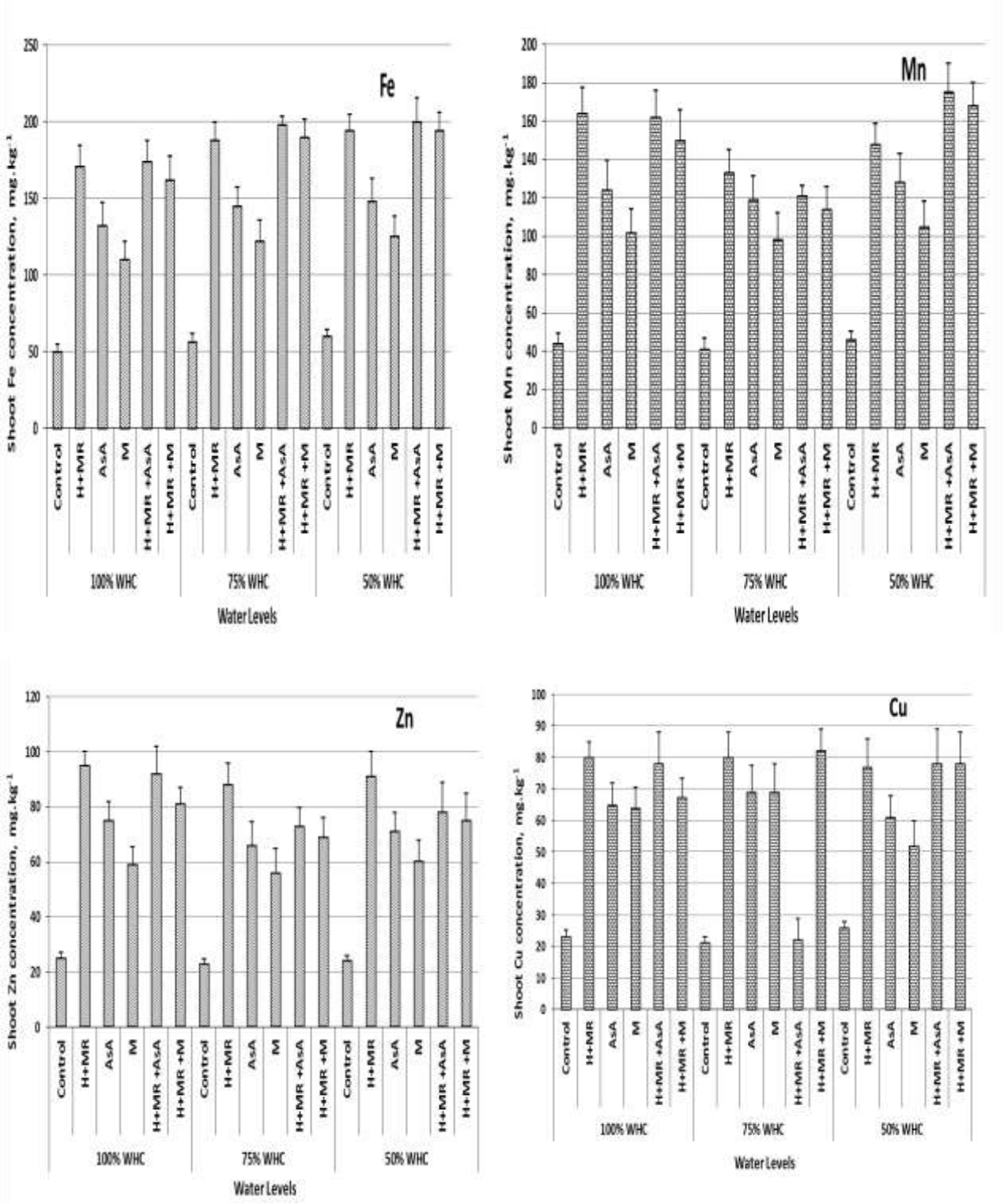

Figure 4. Effects of water stress levels, biofertilization, and antioxidant treatments on some micronutrients concentrations in tissues of corn plants. Error bars on all figures represent the standard error of the mean.

Biofertilizers treatments (H+MR), (H+MR+AsA), and (H+MR+M) had a significant effects on N, P and K concentrations in corn plant as compared to salinity affected plants (Figure1). The concentrations of Na and Cl in biofertilizer-treated corn plants were higher than those of salinity treated plants without biofertilizers except with S3 treatments. On the other hand, application of either ascorbic acid or mannitol alone treatments significantly increased N content, but had no effect on P, and K concentrations in corn plants in comparison to only saline treated plants. Data showed that there is no significant difference between mannitol and ascorbic acid treatments (Fig.1). On the other hand, application of ascorbic acid significantly decreased Na and Cl concentrations in corn plants treated by S3 as compared to control. The combined addition of biofertilizers and ascorbic acid (H+MR+AsA) significantly increased the content of N, P, and K in corn plants compared to control (Fig.1). However, the combination of biofertilizers and ascorbic acid did protect plants against the increase of Na and Cl in corn plants compared to the concentrations of those elements in control or S3 treated plants (Fig.1). Similarly, Biofertilizers treatments either alone or combined with antioxidants (H+MR) ,(H+MR+AsA), and (H+MR+M) had significant effects on N, P and K concentrations in corn plant as compared with water-deficit treated plants (Fig.2). It is evidenced that both antioxidants did not show any effects on the concentration of these macro elements compared to control plants.

Figures (3 and 4) demonstrate the effects of different tested factors on some micronutrients in corn plants. For different salinity levels (S2 and S3), it is recorded that Fe, Zn, Mn, and Cu were significantly decreased with the increase of salinity levels as compared to control. Figure (3) showed that salt stress caused significant decrease in these micronutrients content of corn plants in comparison of control. It is found also that the addition of biofertilizers (H+MR), (H+MR+AsA), and (H+MR+M) significantly increased micronutrients content of corn plants in comparison to salinity treated plants. Also, foliar application of ascorbic acid (AsA) and mannitol (M) significantly increased micronutrients concentrations in corn plants in comparison to salinity treated plants (Fig.3). It can be concluded that, the combined treatments of biofertilizers and ascorbic acid or mannitol significantly ameliorated the negative effects of salinity levels on micro-nutrients concentrations in corn plants. However, (H+MR+AsA) treatment was the best followed by (H+MR+M), then (H+MR). Similarly, at all water-deficit treatments, the combined treatments of biofertilizers and ascorbic acid or mannitol significantly ameliorated the negative effects of water-deficit levels on micronutrients concentrations in corn plants (Fig.4). However, (H+MR+AsA) treatment was the best followed by (H+MR+M) treatment, then (H+MR), while the best results were noticed at 75 % WHC (Figure 4).

These results may be due to some nutritional disturbances that are expected under saline conditions, resulting in high ratios of Na /Ca and Na/K. In the presence of excess NaCl in the growth medium, Na and Cl are accumulated in plant organs, and these ions can affect other mineral elements uptake through competition of membranes which causes nutrient deficiencies in plants (Bohra and Doffling 1993). It is clear, therefore, that salt stress had caused ion imbalance in corn plant. In General, it was expected that all plants may show a level of mycorrhizal infection as soil was not sterilized. However, to confirm that these effects are due to mycorrhizal influence, roots of biofertilizers-treated plants were found to significantly have a higher infection percentage (50-80% range) than roots of untreated corn plants (15-30% range) (data not shown).

CONCLUSION

This study showed the role of antioxidant that improve growth and nutrients content of corn plants grown under moderate to high salinity and/or water-deficit stresses. Inoculation with PGPR (Halix) and Mycorrhizal fungi as biofertilizers is a useful tool for improving nutrients status in tissues of corn plants grown under saline or water-deficit conditions. It can be concluded that the interaction effects of biofertilizers and ascorbic acid or mannitol is a promising approach to promote growth and yield of crops in salt affected soils.

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الملخص العربي

التأثير الحفاز للأسمدة الحيوية ومضادات الأكسدة على الحالة الغذائية ونمو نبات الذرة الشامية تحت تأثير الأجهاد المائى والملحى

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أجريت هذة الدراسة فى الصوبة الزراعية الخاصة بمعمل بحوث الأراضى الملحية والقلوية بأبيس– مركز البحوث الزراعية– وزارة الزراعة واستصلاح الأراضى بهدف تقييم تأثير الرى بمياة مالحة والتعرض للأجهاد المائى على محصول نبات الذرة بالأضافة الى تقليل التغيرات الفسيولوجية والبيوكيميائية التى تحدث بسبب ضغط الملوحة ونقص الماء عن طريق التسميد الحيوى ومضادات الأكسدة. وقد بينت النتائج الى أن زيادة الملوحة قد أدت الى حدوث انخفاض معنوى فى نمو نبات الذرة(الوزن الجاف للساق والجذور) مقارنة بالمعاملات غير المتأثرة بالملوحة.كما بينت النتائج أن المعاملات المزدوجة بكلا من السماد الحيوى (الهاليكس والميكوريزا) ومضاد الأكسدة (حامض الأسكوربيك والمانيتول) لة تأثير معنوى على تقليل الأثر السلبى للملوحة على نمو الذرة.

أدى هذا التأثير المتداخل الى حدوث زيادة معنوية فى محتوى الكلوروفيل داخل أنسجة النبات وكذلك حدوث نقص معنوي فى محتوى حمض الأسكوربيك داخل أنسجة النبات مقارنة بالنباتات غير المعاملة. وقد أدت زيادة مستويات الملوحة فى ماء الرى الى ارتفاع تركيز البرولين داخل أنسجة النبات. وبصفة عامة فأن التأثير المتداخل بين السماد الحيوى (الهاليكس والميكوريزا) ومضاد الأكسدة (حامض الأسكوربيك والمانيتول) أدى الى تحسين معنوى فى تركيزات العناصرالغذائية الصغرى والكبرى داخل أنسجة النبات مما يعادل أويقلل من الأثر السلبى للملوحة.